# Use of supplementary tryptophan to modify the behavior of pigs<sup>1</sup>

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**ABSTRACT:** Three experiments were conducted to investigate the short-term use of supplementary Trp on the behavior of grow/finish pigs. Three levels of dietary Trp were used, representing the standard requirement for growth (control), twice  $(2\times)$ , and 4 times  $(4\times)$ the control amount. In Exp. 1, pigs were fed the diets for 7 d, during which observations were made of their general behavior (time budget), aggression within the group of familiar pigs, and response to a startling auditory stimulus. Behavior effects were evident during the period of supplementation for both the  $2\times$  and  $4\times$  diets. During the treatment period, pigs fed supplemental Trp spent more time lying (P = 0.04) and less time eating (P = 0.05) than pigs fed the control diet. Although the response of the animals to the startling stimulus was to become alert and stand, similar behavioral effects caused by supplemental Trp also were evident after the startling stimulus (P < 0.01). Based on these observations, the subsequent studies retained the same dietary levels of Trp and incorporated a 3-d feeding of diets before behavior testing. In Exp. 2, pigs were fed the experimental diets for 3 d before being regrouped with

unfamiliar pigs on the same diet. Subsequent aggression was affected by Trp supplementation, in that high levels of dietary Trp decreased the total duration of fighting by approximately 50% (P = 0.03). Supplemental Trp had no effect on the number of fights, and there were no differences between the 2 levels of supplemental Trp on any behavior. In Exp. 3, pigs were exposed to specific handling tests on the farm and meat quality assessments after being fed the experimental diets for 3 d. There were no differences among dietary treatments for any of the meat quality characteristic variables measured. The only behavioral or physiological difference observed among the treatments was a slower movement of pigs fed the 4×Trp treatment than control or  $2 \times \text{Trp-fed pigs}$  in a minimal-forced situation (P = 0.04). Response to confinement on a scale, an electric prod, and movement in general did not differ among treatments. High levels of Trp may result in animals avoiding stressful situations if possible, but they seem to have no effect on responses to stressors that animals may experience in a forced situation.

Key words: behavior, handling, pig, tryptophan

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# **INTRODUCTION**

As a limiting essential amino acid, Trp is typically supplied in the diet at levels required for maximum animal growth. When excess Trp is supplied to the diet and not used for the purpose of protein synthesis, it may be used as a therapeutic supplement. The rationale for the therapeutic use of Trp is based on the fact that alterations in brain Trp levels can influence the synthe-

sis of serotonin, an inhibitory neurotransmitter in the central nervous system (**CNS**). Tryptophan is the primary precursor of serotonin. Serotonin has a sedative effect, such as suppressing sleep-wake mechanisms, temperature regulation, pain sensitivity, and aggressive behaviors. As serotonin does not cross the bloodbrain barrier, its effects within the CNS depend on the transfer of Trp across that barrier. Once within the CNS, Trp is readily converted to serotonin. The enzyme Trp hydroxylase is only about half-saturated with its substrate, such that an increase in Trp availability can nearly double the rate of serotonin synthesis (Carlson, 1977; Heine et al., 1995; Sainio et al., 1996).

In domestic animals, therapeutic functions of Trp include decreasing feed intake, modifying aggression, suppressing hysteria, and inhibiting the response to stress (Laycock and Ball, 1990; Chung et al., 1991; Shea et al., 1991; Adeola and Ball, 1992). In swine production

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Table 1. Composition of experimental diets (as-fed basis)

Ingredient	Control	$2\times$	4×
	-	(%)	
Corn	80.43	80.33	80.13
Corn gluten meal	11.68	11.68	11.68
Dicalcium phosphate	2.52	2.52	2.52
Limestone	1.00	1.00	1.00
Vitamin-mineral premix <sup>1</sup>	1.00	1.00	1.00
NaCl	0.40	0.40	0.40
Control premix	2.97	_	_
2× premix	_	3.07	_
4× premix	_	_	3.27
Calculated composition <sup>2</sup>			
Total Trp, %	0.11	0.23	0.43

 $^1\mathrm{Minerals}$  provided the following per kilogram of premix: 10 g of Cu, 16 g of Fe, 5 g of Mn, 20 g of Zn, 100 mg of I, and 20 mg of Se. Vitamins provided the following per kilogram of premix: 1,650,000 IU of vitamin A, 165,000 IU of vitamin D<sub>3</sub>, 8,000 IU of vitamin E, 800 mg of menadione, 1,000 mg of riboflavin, 3,000 mg of D-pantothenic acid, 7,000 mg of niacin, 5 mg of vitamin B<sub>12</sub>, 40 mg of D-biotin, 400 mg of folic acid, 3.31 mg of pyridoxine, and 200 mg of thiamin.

 $^2$ Diets were formulated to contain 3,325 kcal of DE/kg, 13.7% CP, 0.78% total Lys, 0.55% Ca, and 0.17% available P. Percentages of Trp were 0.11, 0.23, and 0.43 for control, 2×, and 4×, respectively.

units and slaughter facilities, there are potentially high levels of stress as animals are moved from their home pens, regrouped, transported, and handled in the packing plant. Suppression of the stress response and associated aggression may be beneficial in terms of animal welfare and meat quality. The present experiments were conducted to determine the effect of short-term supplementation of Trp on behavioral and physiological responses and meat quality indices of pigs within a stable social group, when regrouped, and during handling.

## **MATERIALS AND METHODS**

All experiments were carried out in accordance with the Animal Care Protocol issued by the University of Saskatchewan Committee on Animal Care and Supply.

# Experiment 1

One hundred pigs (Cambrough 15  $\times$  Canabrid) from the Prairie Swine Centre, Inc. in Saskatoon, SK, Canada, averaging 70 kg at the initiation of the study, were assigned randomly to 20 pens of 5 pigs each, segregated by sex (10 pens of each). The pigs were deemed to be in excellent health. Pens were 1.8 m  $\times$  2.4 m, fully slatted, and equipped with one single space feeder and one nipple drinker. Pen walls were solid polyvinyl chloride (**PVC**) panels. The 20 pens were in one negatively ventilated room with end-wall fans and ceiling inlets.

The control diet was a corn-corn gluten meal-based diet and was balanced to meet NRC (1998) nutrient requirements for pigs of this size (3,300 kcal/kg), including 13.7% CP and 0.11% Trp (as-fed basis; Table 1). The experimental diets,  $2\times$  and  $4\times$  (dietary Trp levels

at 2 and 4 times the standard requirement for growth, respectively), were similar to the control diet except that crystalline L-Trp (98% L-Trp; BioKyowa Inc., Chesterfield, MO) was supplemented to achieve 0.23 and 0.43% Trp, respectively. In this and subsequent experiments, "high" tryptophan diets refer to both 2× and 4× treatments unless further qualified. All pens were fed control diets for 7 d. During the subsequent 7 d, pens were assigned randomly within sex to the 3 diets, such that 6 pens were fed control, 7 pens were fed 2×, and 7 pens were fed 4×. All pens were subsequently returned to the control diet for an additional 7 d. Feed was added, weighed back, and changed at approximately 1100 when necessary.

Behavioral observations were made during the mornings of d -4, -3, 0, 1, 2, 3, 4, 7, 8, 14, and 15, where d 0 represented the day that dietary treatments were imposed. Observations were conducted before feed additions, weighbacks, and changes were performed. Five trained observers, blind to the treatments, rotated among 10 observation stations, each consisting of 2 adjacent pens. Observers watched the 2 adjacent pens simultaneously, for periods of 5 min, before rotating to another pair of pens. The proportions of time spent lying, sitting, standing, eating, and drinking were determined by instantaneous scan sampling of pig activity at 1-min intervals (Martin and Bateson, 1993). During 2 complete rotations among the pens, each pen was observed for a total of 50 min (50 scans). During the observations used for determining time budgets, continuous behavior sampling was conducted for frequency and duration of aggression during the 4 min between the first and last scan sample of each 5-min observation period (Martin and Bateson, 1993). This represented 40 min of continuous observation ( $10 \times 4$ -min periods) per pen daily. Aggression was defined as open-mouthed contact with another pig, and a bout of aggression was deemed to have ended when pigs separated for a period of ≥5 s. Beginning on d 0 and continuing on each subsequent observation day, the response of pigs to a frightening stimulus was determined during an additional round of observations. Base activity levels were determined during the initial 10 min (5 min per pen) in the same manner described earlier for time budgets. The animals were then startled by the simultaneous blasting of 4 air horns within the room. A complete round of observations followed immediately, lasting 50 min, with 25 min (25 scans) of observations per pen.

The experimental unit was the pen. Data were classified as pretreatment (d-4, -3, and 0), during treatment (d 1, 2, 3, 4, and 7), and posttreatment (d 8, 14, and 15) periods. Within each period, data were averaged to obtain a single value for each pen. Data were then analyzed within these periods (3 analyses) for the effects of treatment, sex, and the treatment  $\times$  sex interaction tested against pen (treatment sex) using the GLM procedure of SAS (SAS Inst., Inc., Cary, NC).

# Experiment 2

In each of 2 identical trials, 120 pigs (Cambrough 15  $\times$  Canabrid) were allotted to 6 (2.8 m  $\times$  4 m) pens. The pigs were deemed to be in excellent health. Following a 2-wk social adjustment period during which all pigs were fed the control diet, each of the 3 dietary treatments (Table 1) was imposed on 2 randomly allotted pens of pigs for 4 consecutive days. Tests on aggression were conducted on the final 2 d of the dietary treatment. The average BW of the pigs at the time of the aggression tests was  $29.2 \pm 3.6$  kg in Trial 1 and  $27.3 \pm 4.9$  kg in Trial 2. Aggression tests were conducted in a separate room equipped with 20 pens (1.8 m  $\times$  2.4 m). The pens were fully slatted with solid PVC walls. The room was negatively ventilated with end-wall fans and ceiling inlets.

The test situation consisted of a pair of unfamiliar pigs, of opposite sex, that had been fed the same experimental diet. Six pairs, 2 from each dietary treatment, were tested simultaneously in adjacent pens and were referred to as a block. Four blocks of testing were conducted each day. Testing was conducted on 2 d in each of 2 trials. In total, 192 pigs were involved in the testing, in 96 pairs, with 32 pairs per dietary treatment. Two blocks were conducted within the room between washings, so that no tests were conducted in unwashed pens. Once tested, pigs were moved to a third room to prevent contact with untested pigs. Pigs were maintained in familiar groups when moved to the test room and were regrouped as quickly as possible (<5 min) at the beginning of the test.

Aggressive behaviors of the pigs were recorded for 1 h after regrouping. Six trained observers rotated among the pens at 10-min intervals, observing the pen continuously during the interval between rotations. Thus, each pen was observed continuously for 60 min. Observers recorded the frequency and duration of behaviors of interest within each pen. The observed behaviors included the following:

- Parallel pressing (an aggressive behavior): the pigs stand side by side and push hard with the shoulders against each other, throwing the head against the neck or head of the other,
- Inverse parallel pressing (an aggressive behavior):
  the pigs face front to front and then push their
  shoulders hard against each other, throwing the
  head against the neck and flanks of the other,
- Parallel pressing-biting (an aggressive behavior): as parallel pressing or inverse parallel pressing but with bites directed toward head, ears, or flank of the other pig,
- Mutual bite (an aggressive behavior): a pig delivers a bite that is retaliated with a bite from another pig within 5 s,
- Bite (an aggressive behavior): a pig delivers a knock with the head against the head, neck, or body of the other pig with the mouth open,

- Levering (an aggressive behavior): the pig puts its snout under the body of another pig and lifts it up in the air,
- Head-to-body knocking (an aggressive behavior): a rapid thrust upward or sideways with the head or snout against any part of the body behind the ears; most of the knocks are performed against the front half of the receiver, and the performer's mouth is shut.
- Head-to-head knocking (an aggressive behavior): a rapid thrust upward or sideways with the head or snout against the neck, head, or ears of the other; the performer's mouth is shut,
- Nose-to-head contact (start to aggressive behavior or individual recognition): the nose of a pig approaches and contacts the head of another pig and is then withdrawn; if this happens within the context of the delivery of a bite, it will not be recorded as a nose contact, and
- Nose-to-body contact (start to aggressive behavior or individual recognition): the nose of a pig approaches and contacts any part of the body behind the ears, apart from the genital region, of another pig.

Data were analyzed using the GLM procedure of SAS (SAS Inst., Inc.), with each pair of regrouped pigs as an experimental unit. Trial, day within trial, block within day, and their interactions were included in the model. Treatment and all interactions of trial, day within trial, and block within day with treatment were tested against the residual term (df = 48).

#### Experiment 3

One hundred twenty pigs (Cambrough 15×Canabrid) were weighed and assigned to 20 pens of 6 pigs each at approximately 140 to 150 d of age. The pigs were deemed to be in excellent health. There were 6 pens of males and 14 pens of females. The 1.8-m × 2.4-m pens were equipped with fully slatted floors, a single-space dry feeder, and a nipple drinker. Following 2 wk on the control diet, 6 pens (2 pens of males and 4 pens of females) were assigned to each of the 3 dietary treatments (control, 2×, and 4×; Table 1) for 4 d.

Meat Quality. After 3 d on the experimental diets, 2 or 3 larger pigs from each pen were selected for marketing, such that a total of 16 pigs (6 males and 10 females) for each treatment group were obtained. Pigs were tattooed in their home pen and moved to the loading area pen by pen, taking approximately 1 min to move a pen to the loading area. Within the loading area, the pigs were held in a common pen. Animals were loaded onto a truck with the use of electric prods and transported approximately 40 km to a slaughter facility. All pigs received one short electric shock while being loaded to provide a base level of stressful handling. Additional shocks were given at the discretion of the loading and unloading crews as per normal handling

procedures. The pigs were slaughtered after unloading without rest. The total time between moving pigs from their home pens to slaughter was approximately 1 h. These handling procedures were employed to induce a relatively high incidence of PSE and, thereby, provide a strong test of the treatments.

After slaughter, carcasses were chilled at 2°C for approximately 24 h, after which a 10-cm sample was collected from the middle of the loin and cut into 2.5-cmthick chops. Ultimate pH and color were determined, in duplicate, at 24 h postmortem. Ultimate pH was measured with a pH meter (Accumet pH meter 910; Fisher Scientific, Edmonton, Alberta, Canada) fitted with a pear-tip glass probe (Sealed Ag/Agcl; Cole-Parmer, Vernon Hills, IL). Color was determined after a 30-min bloom period (30 min after cutting) by measuring Hunter L\*, a\*, and b\* with a Minolta Chroma Meter (Portable CR-200b 212410390 Colorimeter, Osaka, Japan; Hill et al., 1998). In addition, duplicate standard samples of approximately 50 g were taken from the center of the LM for measurement of 48-h drip loss (Kauffman et al., 1986). Muscle samples were weighed, and 2 filter papers (P5, 5.5-cm diameter, Cat. No. 09-801K; Fisher Scientific) were placed on their transactional surfaces. Samples were then lightly wrapped in a paper towel and individually sealed in air-inflated, zip-polyester bags and subsequently stored at 0°C for 48 h and weighed. Drip loss was determined by calculating total weight loss as a percentage of initial sample weight. Pigs were classified as PSE if their drip loss was >5% and Hunter L\* was >58 (Kauffman et al., 1993).

Handling. The handling test was conducted on nonmarketed pigs in 4 blocks after the third day of the dietary treatments. Within each block, 4 pigs on each treatment from different pens were tested for their response to routine handling, such as moving, weighing, isolation, and regrouping. Before being handled, skin surface temperature and heart rate were determined in their home pens. Skin temperature was measured by an infrared thermometer (Oakton Infrapro 3; Cole-Parmer) at 2 locations, behind the ear and on the rump. Polar heart rate monitors (Polar Beat HRM; Polar Electro Oy, Kempele, Finland), designed to monitor heart rate during human exercise, were used to determine the heart rate. These monitors consisted of 2 electrical sensors attached to an elastic belt, strapped around the thorax of the pig. Input to these sensors was integrated to determine heart rate, which was transmitted a short distance to a hand-held monitor and recorded. Following the prehandling measurements, the pigs were moved from their home pens to another test room. The individual pigs were brought to a start point at the exit of their home room and then herded, without prodding, to the end point, close to the scale and holding pens. The distance between the start point and the end point was 20 m. The walking time was recorded. The pigs were then weighed on a crate scale and put into individual pens for 5 min of isolation. One-half the pigs from each treatment group received 2 short-duration electric shocks from a prod while in the scale. The  $1.8\text{-m} \times 2.4\text{-}$ m isolation pens had solid walls that prevented pigs from contacting each other. Skin temperatures were measured in the scale immediately before release (after prodding, if applied). Heart rate was determined at the end of the walk, in the scale, immediately before release (after prodding, if applied), and after 5 min of isolation. Four unfamiliar pigs on the same treatment were regrouped in a holding pen for 1 h, during which the behavior of the pigs was videotaped. Immediately following the last pig entering the holding pen, fighting incidence (frequency and duration) was recorded by continuous observation. Meanwhile, lying and exploratory (sniffing, biting, rooting, and chewing walls and floor) behaviors were recorded by instantaneous scan sampling at 5-min intervals.

For the meat quality portion of the experiment, the individual pig was considered the experimental unit. Data were analyzed by one-way ANOVA using GLM. The main factor was dietary Trp level with individual pig as the experimental unit. The incidence of PSE was tested by  $\chi^2$  analysis. For the handling test, a randomized block ANOVA was employed, with individual animal as the experimental unit. For all variables assessed before weighing, the model included dietary Trp treatment only. For the variables measured after weighing, the main factors of dietary Trp and prodding, and their interaction, were tested by the residual error. To test changes in skin temperature and heart rate from prehandling to posthandling, repeated measures analysis of variance was used.

#### **RESULTS**

# Experiment 1

Feed intake during the pretreatment phase was approximately 30% less than during the treatment or posttreatment periods (2.06 vs. 2.73 and 2.71 kg/d, respectively). Males consumed more feed than females during both the treatment and posttreatment periods (2.91 vs. 2.54  $\pm$  0.12 kg/d, P < 0.05; 2.91 vs. 2.51  $\pm$  0.05 kg/d, P < 0.01, respectively), but not during the pretreatment period. At no time did the feed intake by pigs on different dietary treatments differ.

No differences in activity time budgets were observed among dietary treatments during either the pre- or posttreatment periods. During the treatment period, pigs fed high-Trp diets spent less time eating (P=0.05) and more time lying (P=0.04) during the prestartle phase (Table 2). The response of the animals to the startling stimulus was to become alert and stand. Gradually, the animals returned to their previous activity over the course of the observations. Subsequent to being startled, those pigs fed high-Trp diets continued to spend more time lying (P=0.01) and also spent less time standing (P=0.01) than the pigs fed the control diet. Neither the frequency nor the duration of aggression differed among dietary treatments during any of the 3 periods of the study.

**Table 2.** Activity time budgets (percentage of time) before and after startling stimulus during treatment period for pigs fed different levels of Trp in Exp. 1

Time		D	Dietary Trp level $^1$			
	Activity	Control $(n = 6)^2$	2× (n = 7)	4× (n = 7)	SEM	<i>P</i> -value
Pre-startle						
	Sitting	3.8	3.6	4.7	1.1	0.76
	Drinking	2.6	2.5	2.3	0.3	0.75
	Eating	$14.1^{x}$	$10.4^{\mathrm{y}}$	$10.5^{y}$	1.1	0.05
	Lying	$58.2^{y}$	$66.6^{x}$	$69.3^{x}$	3.0	0.04
	Standing	21.5	17.0	13.3	2.6	0.11
Post-startle						
	Eating	14.1	13.6	13.3	0.9	0.83
	Lying	$52.3^{\mathrm{y}}$	$59.8^{x}$	$62.7^{x}$	2.5	0.01
	Standing	$27.0^{x}$	$19.9^{y}$	$16.9^{y}$	1.8	0.01

<sup>&</sup>lt;sup>x,y</sup>Means within rows that do not have a common superscript differ, P < 0.05.

# Experiment 2

Following regrouping, pigs started nosing each other within 1 min. The nosing phase lasted approximately 20 to 30 min before fighting began. The only effect of Trp treatment during the nosing phase was a decrease in the duration of nose-to-head nosing in those pairs fed the  $2\times$  diet (P=0.04; Table 3).

Fighting latency for individual pigs varied widely, from 2 s to >60 min after regrouping, such that there were no significant differences in the mean latency to fight among the treatment and control groups (Table 4). Approximately 20% of pigs did not fight during the first hour of regrouping in either of the treatment or control groups. Pigs involved in fighting usually had one long and intense fight, which was followed by a few short fights.

Fighting among pigs consisted mainly of pressing and mutual biting. Total fighting time was calculated by

summarizing durations of parallel pressing, inverse parallel pressing, and mutual biting. High levels of dietary Trp decreased total duration of fighting by approximately 50% (P=0.03). Although several aggression variables evidenced a numerical decrease in pigs fed the high-Trp treatments, only inverse parallel pressing was significantly decreased. The average duration of a bout of inverse parallel pressing was decreased on both high-Trp treatments (P=0.05), and combined with a numerical decrease in frequency, the total duration of inverse parallel pressing was reduced to approximately one-third that of control (P=0.04). In terms of differences between the 2 supplemented levels of Trp, no significant difference was found in any behavior.

# **Experiment 3**

For the meat quality portion of the experiment, the preslaughter handling resulted in lower pH and higher

**Table 3.** Effects of dietary Trp on nosing behaviors in growing pigs during the initial hour after regrouping in Exp. 2

	D	ietary Trp level	l <sup>1</sup>	SEM	<i>P</i> -value
Item	Control $(n = 32)^2$	2× (n = 32)	4× (n = 32)		
Latency to first nosing, s	22.4	18.6	24.4	8.78	0.90
Duration of nosing phase, min	24.8	22.8	28.4	3.95	0.61
Nose-to-head					
Frequency	34.3	27.2	34.2	2.69	0.11
Total duration, s	$215.1^{xy}$	$170.7^{\mathrm{y}}$	$241.5^{x}$	19.3	0.04
Average duration, s	6.58	7.91	7.22	1.41	0.80
Nose-to-body					
Frequency	15.9	13.6	15.9	1.5	0.46
Total duration, s	104.2	85.8	88.5	10.8	0.44
Average duration, s	6.45	7.21	5.58	0.71	0.27

<sup>&</sup>lt;sup>x,y</sup>Means within rows that do not have a common superscript differ, P < 0.05.

<sup>&</sup>lt;sup>1</sup>Percentages of Trp were 0.11, 0.23, and 0.43 for control, 2×, and 4×, respectively.

<sup>&</sup>lt;sup>2</sup>Number of animals in each pen, which was the experimental unit in this experiment.

<sup>&</sup>lt;sup>1</sup>Percentages of Trp were 0.11, 0.23, and 0.43 for control, 2×, and 4×, respectively.

<sup>&</sup>lt;sup>2</sup>Pairs of animals in each pen, which was the experimental unit in this experiment.

**Table 4.** Effects of dietary Trp on aggressive behaviors in growing pigs during the initial hour after regrouping in Exp. 2

Item	Dietary Trp level <sup>1</sup>				
	Control $(n = 32)^2$	2× (n = 32)	4× (n = 32)	SEM	<i>P</i> -value
Fighting					
Latency, min	25.0	23.2	28.8	4.0	0.60
Total duration, s	$147.2^{x}$	$79.2^{\mathrm{y}}$	$53.1^{y}$	24.9	0.03
Biting					
Frequency	11.9	9.3	8.7	1.5	0.28
Mutual biting frequency	5.3	3.5	2.6	1.0	0.14
Parallel press					
Frequency	2.88	3.06	2.47	0.46	0.65
Total duration, s	22.9	25.8	14.8	4.3	0.19
Average duration, s	6.12	5.48	4.13	0.89	0.28
Inverse press					
Frequency	5.72	3.25	2.44	1.27	0.17
Total duration, s	84.8 <sup>x</sup>	$28.7^{y}$	$19.0^{y}$	18.9	0.04
Average duration, s	7.84 <sup>x</sup>	$4.52^{xy}$	$4.43^{y}$	1.28	0.05

<sup>&</sup>lt;sup>x,y</sup>Means within rows that do not have a common superscript differ, P < 0.05.

drip loss than expected in normal pork (Kauffman et al., 1993). The color scores were not extreme, resulting in only a moderate number of pigs exhibiting PSE (3, 1, and 5 for control,  $2\times$ , and  $4\times$ , respectively). There were no differences among dietary treatments for any of the meat quality characteristics measured (Table 5).

Ear surface temperature decreased (31.33 vs. 30.25; P = 0.02), whereas rump surface temperature increased (29.62 vs. 31.40; P = 0.02; prehandling vs. postweighing,respectively) in response to handling. Heart rate increased (131.7, 158.2, and 164.2 beats/min, prehandling, postexercise, and postweighing, respectively; P =0.01), but it returned to normal (138.2 beats/min) during the 5 min of isolation. Prodding the pigs while in the scale increased their heart rate compared with nonprodded pigs (180.1 vs. 154.3; P = 0.01), but this returned to nonprodded levels by the end of the 5-min isolation period. Prodding had no effect on skin temperatures. Pigs fed the highest level of Trp (4×) took longer to walk from the start point to the scale (P = 0.04). Heart rates and skin temperatures were not affected by dietary Trp levels (Table 6).

## **DISCUSSION**

Pigs are very adaptable to their environment, but nutritional status may affect how they respond to environmental stimuli. To date, however, there has been limited research conducted in the area of Trp nutrition and behavior in swine. In addition, much of it is inconsistent with expected metabolite changes. McGlone et al. (1985) evaluated the effect of 0.5% Trp on regrouping and heat stress in 8.6-kg pigs and reported that regrouped pigs fed Trp had improved ADG and ADFI during the first 7 d of the trial; however, neither regrouping nor Trp alone affected growth. Seve et al. (1991) evaluated brain metabolites and behavior (grunts, squeals, ambulation, and exploration in an

**Table 5.** Effects of dietary Trp on meat quality characteristics of market pigs in Exp. 3

	]	Dietary Trp level <sup>1</sup>	<u> </u>		
Item	Control $(n = 16)^2$	2× (n = 16)	4× (n = 16)	SEM	<i>P</i> -value
Moving to loadout, s Color <sup>3</sup>	59.4	48.9	60.0	5.72	0.33
L	53.3	52.9	53.4	1.2	0.96
a*	9.1	8.0	8.7	0.36	0.11
b*	4.2	3.4	4.1	0.44	0.38
24-h pH	5.23	5.31	5.29	0.032	0.18
48-h drip loss, %	15.6	15.1	14.2	1.3	0.75

<sup>&</sup>lt;sup>1</sup>Percentages of Trp were 0.11, 0.23, and 0.43 for control, 2×, and 4×, respectively.

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<sup>&</sup>lt;sup>3</sup>Determined 30 min after cutting by measuring Hunter L\*, a\*, and b\* with a Minolta Chroma Meter (Portable CR-200b 212410390 Colorimeter, Osaka, Japan; Hill et al., 1998).

**Table 6.** Effects of dietary Trp on responses to handling in market pigs in Exp. 3

Item	D	ietary Trp level	1	SEM	
	Control $(n = 16)^2$	2× (n = 16)	4× (n = 16)		<i>P</i> -value
Movement time, s	78.3 <sup>y</sup>	80.2 <sup>y</sup>	131.7 <sup>x</sup>	16.5	0.04
Heart rates, beats/min					
Prehandling	133.8	127.4	139.3	4.0	0.11
Postexercise	152.1	160.8	170.3	7.0	0.20
Postisolation $(n = 4)$	131.8	145.5	138.3	8.7	0.56
Skin surface temperature, °C					
Ear, prehandling	32.1	31.0	30.7	0.58	0.22
Ear, postexercise	30.9	30.2	30.1	0.46	0.39
Rump, prehandling	31.0	29.1	29.0	0.73	0.12
Rump, postexercise	31.6	31.3	31.4	0.70	0.95

<sup>&</sup>lt;sup>x,y</sup>Means within row that do not have a common superscript differ, P < 0.05.

open-field test on d 5, 23, and 45 after weaning) in weanling pigs given varied levels (0.14, 0.23, or 0.32%) of dietary Trp and reported that behavioral reactivity was not influenced by dietary Trp, even though brain Trp and Trp:large neutral amino acids were increased. In a companion paper, Meunier-Salaün et al. (1991) reported that dietary Trp levels induced large variations in brain amino acids and indole concentrations, but changes in behavioral responses were minor. Adeola and Ball (1992) and Adeola et al. (1993) reported that concentrations of serotonin in the hypothalamus of stress-susceptible pigs were less than in stress-tolerant pigs, even though Trp concentrations did not differ. These studies did not evaluate the effect of dietary Trp on any behavioral variables. A differential response to supplemental Trp, in which aggression was decreased to a greater extent in dominant than in subordinate chickens, was reported by Shea et al. (1991). Recently, Peeters et al. (2004) reported that pigs provided with Trp in their drinking water spent more time lying during simulated transport than control pigs, but no other differences were observed.

In Exp. 1, intakes were low during the week of adaptation to the control diet relative to feed intake the previous week, potentially because of the change from the standard farm diet or the change in environment with movement to the experimental room. Intakes during the experimental period returned to normal and did not differ among treatments. This result suggests that at levels of 0.43% Trp did not have an intake suppressing effect as did higher levels reported in previous studies (Chung et al., 1991; Rosebrough, 1996). The time spent eating was decreased on the high-Trp diets during treatment, although these observations were only for a short portion of the day. More importantly, pigs fed high levels of Trp (both  $2\times$  and  $4\times$ ) spent more time lying and tended to spend less time standing during the treatment period. An increase in lying in response to Trp supplementation also was reported by Peeters et al. (2004). In our study, these differences also

extended into the startle recovery period. The change in behavior on treatment was generally evident on the first observation day after diet change. Behavioral differences disappeared within 3 d of Trp removal. No differences were observed between the 2 levels of Trp supplementation. These results suggest that the levels of supplementation used were effective within a few days of application.

Aggression was studied in all 3 experiments, although it was the principle focus only in Exp 2. The use of familiar social groups in Exp. 1 and the inadequate number of experimental units in Exp. 3 precluded obtaining significant differences among treatments unless dramatic effects were present. The results of Exp. 2 indicated that aggression was decreased in both high-Trp treatments. No effect was found in the preliminary investigative (nosing) phase of the fight, but rather in the intensely aggressive portion. The most severe form of fighting is inverse parallel pressing, where neither pig has gained an advantage and their efforts are intense. This portion of the fight was decreased in the Trp-treated animals. The effect was a decrease in the total duration of fighting and in the intensity of those fights. A similar differential response in aggression to Trp supplementation was reported in mature broiler breeder males, in which threatening behavior was less affected than was actual pecking (Shea et al., 1990).

High levels of dietary Trp, 2× and 4×, slowed the movement of pigs in a minimal-force handling situation; however, the same pigs responded to weighing and electrical shock to the same degree as pigs fed the control diet. The response of pigs to high levels of dietary Trp may be situation-specific. When pigs are in a situation that they control, such as resting in a pen, when choosing to terminate a fight, or when handled with minimal force, they were less excitable when fed high levels of Trp. When stress was unavoidable, such as with confinement in a scale or receiving an electric shock, pigs responded similarly regardless of the dietary Trp level. This result is similar to that of Meunier-Salaün et al.

<sup>&</sup>lt;sup>1</sup>Percentages of Trp were 0.11, 0.23, and 0.43 for control, 2×, and 4×, respectively.

<sup>&</sup>lt;sup>2</sup>Number of animals in each pen, which was the experimental unit in this experiment.

(1991), who reported that dietary Trp could induce large variations in brain serotonin levels, but minor changes in behavioral or cortisol responses to stress. Peeters et al. (2004) reported no differences in responses to simulated transport in Trp-supplemented pigs, with the exception that they resumed lying earlier than did control pigs, somewhat similar to our pigs' recovery from the startle application.

Because of the metabolic relationships between dietary Trp and serotonin, behavior type and serotonin production, stress at slaughter and hypothalamic neurotransmitter concentration, and stress at slaughter and the production of PSE pork, it could be hypothesized that supplemental Trp may help alleviate the negative effects of stress at slaughter on meat quality. Meat quality, however, is complex, as there are many contributing factors including environment, genetics, and plane of nutrition (Forrest et al., 1975). Adeola and Ball (1992) evaluated the use of supplemental Trp in 92kg pigs and reported that Trp supplementation lowered the incidence of PSE pork compared with pigs fed the control diet (27 vs. 33%, respectively); however, the pH, color, and structure of the loins and hams were similar regardless of the diet fed. Henry et al. (1992) fed 44- to 90-kg pigs a diet deficient in Trp and reported that pigs fed the Trp-deficient diet had higher initial pH levels in the ham and loin. In turkeys, Denbow et al. (1993) tested the effect of 0.1, 0.2, and 0.3% supplemental Trp for 2 wk before market and reported that dietary Trp had no effect on muscle shear, color, or pH (0, 4, or 24 h), despite changes in brain metabolite concentrations. Although we did not measure plasma Trp or its metabolites, we did not see any effect of supplemental Trp on meat quality characteristics.

There are 2 possibilities that may explain the lack of differences in meat quality among the treatments. The first is that our handling regimen was too severe and overwhelmed any possibility of differential response. Specifically, the animals were rested for a shorter period of time at the packing plant than normal; however, this resting time relates to metabolic recovery from the stress response and not to a decrease in the stress itself. In other words, the results suggest that all pigs reacted similarly to the stressful conduction. Our hypothesis was that high-Trp diets would decrease the reaction of animals to these stressors, not that it would improve their recovery. The second possibility is that Trp is effective in nonforced or animal-controlled situations. In Exp. 2, the length of a fight was decreased, as the fight itself and its length were under the control of the animal. In addition, under nonforced movement, high-Trp pigs moved more slowly. When movement was forced (moving to loadout), they moved as quickly as the control pigs. Similarly, when animals had an electric shock forced on them, all physiological responses were similar across diets.

In conclusion, behavioral effects were evident in pigs within a few days of being fed diets that provided Trp at  $2\times$  and  $4\times$ . These behavioral differences included a

greater time spent lying and less time spent eating. Aggression among unfamiliar pigs was decreased in duration and intensity, but not frequency, when pigs were fed high-Trp diets. The responses of pigs to the stressors of handling, including electric shock, were unaffected by Trp treatment. Short periods of high dietary levels of Trp could be used to decrease aggression, but high levels of Trp seem to have little effect on the response to handling. Therefore, it is suggested that high levels of Trp may result in avoidance of stressful situations, if possible. However, these higher levels of Trp have no effect on response to stressors that are forced on the animal. High dietary Trp had no effect on the color, pH, or drip loss of the LM.

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